Animal: An International Journal of Animal Bioscience

Light-based monitoring devices to assess range use by laying hens

--Manuscript Draft--

Manuscript Number:	ANIMAL-19-40538R3
Full Title:	Light-based monitoring devices to assess range use by laying hens
Short Title:	Light-based monitoring devices to assess range use
Article Type:	Research Article
Section/Category:	4a. Welfare and Behaviour
Keywords:	poultry; range use; outdoor; automatic monitoring; cover
Corresponding Author:	Stephanie Buijs UNITED KINGDOM
First Author:	Stephanie Buijs
Order of Authors:	Stephanie Buijs
	Christine J Nicol
	Francesca Booth
	Gemma Richards
	John F Tarlton
Manuscript Region of Origin:	UNITED KINGDOM
Abstract:	Access to an outdoor range has many potential benefits for laying hens but range use can be poor due to factors only partly understood. Techniques to monitor individual range use within commercial flocks are crucial to increase our understanding of these factors. Direct observation of individual range use is difficult and time-consuming, and automatic monitoring currently relies on equipment that is difficult to use in an on-farm setting without itself influencing range use. We evaluated the performance of a novel small, light, and readily portable light-based monitoring system by validating its output against direct observations. Six commercial houses (2000 hens/house) and their adjacent ranges were used, three of which were equipped with more structures on the range than the others (to determine whether cover would influence monitoring accuracy). In each house 14 hens were equipped with light monitoring devices for 5 discrete monitoring cycles of 7-8 consecutive days (at 20, 26, 32, 36 and 41 weeks of age). Light levels were determined each minute: if the reading on the hen-mounted device exceeded indoor light levels the hen was classified as outside. Focal hens were observed directly for 5 minutes/hen/week. Accuracy (% of samples where monitoring and direct observations were in agreement) was high both for ranges with more and with fewer structures, although slightly better for the latter (92 vs. 96% ± 1 SEM, $F_{1,19}=5.2$, $P=0.034$). Furthermore, accuracy increased over time (89, 94, 95, 98% ± 1 SEM for observations at 26, 32, 36 and 41 weeks, respectively, $F_{3,19}=3.2$, $P=0.047$), probably due to progressively reduced indoor light levels resulting from partial closing of ventilation openings to sustain indoor temperature. Light-based monitoring was sufficiently accurate to indicate a tendency for a greater percentage of monitored time spent outside when more range structures were provided (more: 67%, fewer: 56%, SEM: 4, $\chi^2_1=2.9$, $P=0.089$). Furthermore, clear and relatively consistent individual differences were detected. Individuals that were caught outside at the start of the experiment ranged more throughout its duration (caught outside: 72%, caught inside 51%, SEM: 4, $\chi^2_1=10.0$, $P=0.002$), and individual range use was correlated between monitoring cycles (for adjacent monitoring cycles: $r_s^2=0.5$-0.7, $P<0.0001$). This emphasizes the importance of studying range use on an individual level. In conclusion, our light-based monitoring system can assess individual range use accurately (although accuracy was affected by house characteristics to some extent) and was used to show that both cover availability and individual characteristics affected range use.

Manuscript

Light-based monitoring devices to assess range use by laying hens

S. Buijs[1,2], C.J. Nicol[3], F. Booth[1], G. Richards[1] and J.F. Tarlton[1]

[1] School of Veterinary Sciences, University of Bristol, Langford house, Langford, BS40 5DU, United Kingdom

[2] Agri-Food and Biosciences Institute, Large Park, Hillsborough, BT26 6DR, United Kingdom

[3] Royal Veterinary College, Hawkshead Lane, Hatfield, AL9 7TA, United Kingdom

Corresponding author: Stephanie Buijs

E-mail: stephanie.buijs@afbini.gov.uk

Received 20 May 2019; Accepted 12 September 2019

Short title

Light-based monitoring devices to assess range use

Abstract

Access to an outdoor range has many potential benefits for laying hens but range use can be poor due to factors only partly understood. Techniques to monitor individual range use within commercial flocks are crucial to increase our understanding of these factors. Direct observation of individual range use is difficult and time-consuming, and automatic monitoring currently relies on equipment that is difficult to use in an on-farm setting without itself influencing range use. We evaluated the performance of a novel small, light, and readily portable light-based monitoring system by validating its output against direct observations. Six commercial houses (2000 hens/house) and their adjacent ranges were used, three of which were equipped with more structures on the range than the others (to determine whether cover would influence monitoring accuracy). In each house 14 hens were equipped with light monitoring devices for 5 discrete monitoring cycles of 7-8 consecutive days (at 20, 26, 32, 36 and 41 weeks of age). Light levels were determined each minute: if the reading on the hen-mounted device exceeded indoor light levels the hen was classified as outside. Focal hens were observed directly for 5 minutes/hen/week. Accuracy (% of samples where monitoring and direct observations were in agreement) was high both for ranges with more and with fewer structures, although slightly better for the latter (92 vs. 96% ± 1 SEM, $F_{1,19}$=5.2, P=0.034). Furthermore, accuracy increased over time (89, 94, 95, 98% ± 1 SEM for observations at 26, 32, 36 and 41 weeks, respectively, $F_{3,19}$=3.2, P=0.047), probably due to progressively reduced indoor light levels resulting from partial closing of ventilation openings to sustain indoor temperature. Light-based monitoring was sufficiently accurate to indicate a tendency for a greater percentage of monitored time spent outside when more range structures were provided (more: 67%, fewer:

56%, SEM: 4, χ^2_1=2.9, P=0.089). Furthermore, clear and relatively consistent individual differences were detected. Individuals that were caught outside at the start of the experiment ranged more throughout its duration (caught outside: 72%, caught inside 51%, SEM: 4, χ^2_1=10.0, P=0.002), and individual range use was correlated between monitoring cycles (for adjacent monitoring cycles: r_s^2=0.5-0.7, P<0.0001). This emphasizes the importance of studying range use on an individual level. In conclusion, our light-based monitoring system can assess individual range use accurately (although accuracy was affected by house characteristics to some extent) and was used to show that both cover availability and individual characteristics affected range use.

Keywords: poultry, range use, outdoor, automatic monitoring, cover

Implications

A novel light-based monitoring system was shown to provide accurate information on the time individual laying hens spend outside. The system was used to show that hens tended to spend more time outside if there were more structures on their range, and indicated clear differences between individuals within the same flock that remained relatively constant throughout the laying period. This emphasises the importance of studying range use on an individual level.

Introduction

Access to an outdoor range can improve several aspects of laying hen welfare (Knierim, 2006). Apart from providing a preferred environment for foraging and dustbathing (Campbell *et al.* 2017a), associations between increased ranging and a reduction in important welfare problems have been reported (feather pecking: Lambton *et al.* 2010, feather damage: Bestman and Wagenaar, 2003; Nicol *et al.* 2003; Mahboub *et al.* 2004, fearfulness: Campbell *et al.* 2016; Hartcher *et al.* 2016, keel bone fractures: Richards *et al.* 2012), even though cause and effect are often difficult to distinguish. Range use is only one of several factors influencing these welfare problems, as emphasized by other studies that did not find significant associations with range use (feather pecking: Gilani *et al.* 2014; Hartcher *et al.* 2016; fearfulness: Mahboub *et al.* 2004). Therefore, accurate methods of assessing range use are crucial when determining how it contributes to welfare.

The simplest way of assessing range use is by human observation (either directly or by video or photo surveillance). Because of the low set up cost and the ease of application in different settings this remains a popular method (e.g., Gilani *et al.* 2014; Larsen *et al.* 2017; Pettersson *et al.* 2017). However, observations may be unreliable when ranges are large or contain structures obscuring hens from sight, or when observations do not cover all relevant times of the day (as range use changes throughout the day, Bubier and Bradshaw, 1998; Dawkins *et al.* 2003; Chielo *et al.* 2016). Crucially, it is an extremely time-consuming method unless limited to generating flock-level data. This has led the majority of previous studies to focus on flock-level range use, without distinguishing between situations where all hens range at a medium frequency and situations where some hens use the range very frequently whilst others use it very infrequently. However, more recent studies indicate that individual range use differs greatly within a flock (Campbell *et al.* 2016; Gebhardt-Henrich *et al.* 2014), for reasons that are presently unclear. This means that using individual data is essential to gain understanding of why hens range, especially because the welfare problems associated with poor range use, such as feather damage and keel bone fractures, influence welfare of the affected individuals rather than the entire flock.

Automated monitoring of range use allows highly efficient data collection at the level of the individual. Most often, RFID (radio-frequency identification) technology is used to study laying hens' range use automatically (e.g., Campbell *et al.* 2016; Richards *et al.* 2011). Although RFID systems can register pophole passage very accurately (97-99%, Thurner and Wendl, 2005; Thurner *et al.* 2010), these do have some severe limitations. When hens move through the pophole at speeds above 5.4 km/h certain RFID systems are less likely to register them, which distorts ranging data considerably (Gebhardt-Henrich *et al.* 2014). Laying hens often run out when the popholes are opened in the morning and run back in when something frightens them (personal observation). This may lead to undetected ranging bouts especially for quicker or more easily frightened hens, potentially introducing a systematic bias. Also, RFID systems require that each pophole is equipped with sensors covering the full length on both the outdoor and the indoor side (Gebhardt-Henrich *et al.* 2014) or that small popholes are used (Thurner *et al.* 2010; Hartcher *et al.* 2016; Campbell *et al.* 2017b), and often require close proximity to a computer and power supply (Hartcher *et al.* 2016). All of this is unpractical when working on commercial farms, and constraining pophole size to improve accuracy may decrease ranging (Gilani *et al.* 2014). Studying range use on commercial farms is of crucial importance, as research facilities generally only have the capacity to house smaller flocks which show different ranging patterns (Bestman and Wagenaar, 2003; Gilani *et al.* 2014). Ultra-wideband systems can monitor broiler chickens' range use with considerable accuracy (Stadig *et al.*, 2018), but require several elevated receivers that are even more difficult to install rapidly on commercial farms.

As an alternative to RFID and ultra-wideband systems, we developed a system to monitor range use that is quickly and easily set up and moved between farms. This system uses lightweight hen-mounted devices that measure and store light levels without the need to communicate with a receiver. As it is generally considerably darker inside the house than outside on the range, such devices can be used to tell if the hen is outside or inside the house. Lindholm *et al.* (2016) used light monitoring devices to record range use in broiler chicken using fixed threshold values (<125 lux = inside, >300 lux = outside). However, using fixed threshold values may lead to an underestimation of ranging around dusk, a known peak time for ranging (Bubier and Bradshaw, 1998; Dawkins *et al.* 2003) and an overestimation of ranging when sunlight enters the house. Both will decrease accuracy and distort diurnal patterns. To overcome such problems, the system evaluated in the current study compared the light levels recorded by the hen-mounted devices to those of similar devices placed inside and outside the house. This allowed us to continue monitoring under decreased light conditions (e.g. at dusk, or due to bad weather) and to discard data if light levels in certain parts of the house were similar to those outside (i.e., when a considerable amount of sunlight entered the house through ventilation openings). In addition to bright patches inside the house, shaded patches outside can also decrease the accuracy of light-based monitoring. This can be especially problematic because adding cover structures to the range is a popular way of encouraging range use. These structures often cast a shadow (which may partially explain their attractiveness, Nagle and Glatz 2012). Therefore, we tested the accuracy of our light-based monitoring system when applied to ranges with more and fewer cover structures, aiming to determine its accuracy under both conditions. The study spanned several discrete monitoring cycles to determine whether seasonal differences affected accuracy. This could be due to seasonal changes in light levels (both direct and resulting from adjustment of ventilation openings in response to changes in temperature) and hen behaviour (e.g., increased shade use on hot days).

In addition, we evaluated whether our system was sensitive enough to confirm hypotheses based on previous reports. Specifically, we expected that range use would be greater when more cover structures were provided on the range (Hegelund *et al.* 2005; Zeltner and Hirt 2008; Bestman and Wagenaar 2003), and that monitored individuals caught outside prior to the first monitoring cycle of the experiment would range more (Buijs *et al.* 2017). Also, based on the hypothesis that ranging behaviour is driven by long-term individual characteristics, we expected individual range use to correlate between monitoring cycles.

Material and methods

Housing and animals

The experiment was conducted on a commercial farm with six identical houses (Halo Ranger), each housing 2000 British Blacktail hens. Hens had been reared without outdoor access, arrived at the farm when 16 weeks old, and were allowed access to the range two weeks later. The houses (Figure 1) consisted of a slatted area (22.5 × 9 m, raised 1.5 m above the ground where feed, water, perches and nest boxes were available) and a straw covered litter area (20.5 × 9 m) which were connected by a slatted ramp (2 × 9 m). The houses were naturally ventilated through openings of adjustable height along the full length of the house and through the pop-holes. Each house had seven popholes (each 2.5 m wide, with height varying daily between 20 and 50 cm depending on how far the shutters were raised) connecting each house to its own range (approximately 2 ha). To facilitate movement between the indoor and outdoor area slatted ramps were provided outside the popholes of the raised slatted area. Pophole thresholds (present in the litter area only) were low enough for the hen to step onto easily (approximately 15 cm). Houses were oriented north-to-south lengthwise, with the exception of house 6 which was oriented east-to west. The houses were placed centrally at one side of the range (3x north side, 2x south side, 1x east side), providing direct access to the approximately equally sized range areas on either side, as well as indirect access to the area behind the house.

All ranges contained some cover, as required by the farm’s certification scheme: four trampoline-like structures roofed with wind break cloth (1.5 × 1.5 m) and a stack of cut fir trees placed on their sides (approximately 1 × 30 × 0.6 m, Figure 2). Extra cover structures were placed on three out of six ranges, to assess the effect of cover on the accuracy of the monitoring system, as well as on ranging. Each of these ranges contained two tunnel-shaped shelters of corrugated iron (as used in outdoor pig husbandry), and four tent-like structures (3.5 × 2.5 m) and five artificial zig-zag pattern hedges (10 m) made of wind break cloth. These structures were placed on a line extending outwards from the house and to the back of the range. The two types of range will be referred to as the ‘fewer structures’ and ‘more structures’ treatment throughout the paper. On the ‘fewer structures’ ranges the trampoline-like structures were placed further away from the house than on the ‘more structures’ ranges (at 20 and 40 m instead of at 4 m). The treatments were distributed in such a way that the ‘fewer structures’ ranges had a line of trees on one side of the range, whereas the ‘more structures’ ranges had a line of trees and high shrubs on two sides of the range (approximately 3m outside range’s fence).

Light-based monitoring system

Light monitoring devices (Biotrack Ltd., Wareham, UK) were used to measure and store light levels at one minute intervals (except when the devices produced a gap each 17th minute to store the data). Prior to the experiment all devices had been exposed to a standardised light level to calculate normalisation values to remove any individual differences in sensitivity. The devices were mainly sensitive to the blue part of the light spectrum. Such light was emitted from the fluorescent lamps inside the houses in very low amounts and therefore the devices did not pick up the light from these lamps. Devices were mounted on the focal hens and placed in the environment (inside and outside the house). Device placement and data processing are described in more detail below. Briefly, a hen was classified as outside if the reading on the hen-mounted device exceeded the highest reading on any of the indoor devices, except those near the popholes in the slatted area. The readings of the indoor devices near the popholes of the slatted area, and the difference between the levels of the indoor and outdoor devices, were used as thresholds for data inclusion.

Hen-mounted light monitoring devices. In each house, 14 hens were fitted with an approximately 50 g backpack containing a light monitoring device (11.4 g), a commercially available locator device (Tile Mate, Tile Inc., San Mateo, United States) and an accelerometer (Custom Idea Ltd, Shepton Mallet, United Kingdom). The locator device indicated the distance between the hen and a handheld receiver and was only used to aid the detection of the hen prior to direct observations. The accelerometer was not used for the part of the study described here. All equipment was wrapped in brown electrical tape with the tip of the light monitor sticking out to allow light measurement. This package was attached to the hen by elastic loops around the wings (Figure 3). In previous studies (Buijs *et al.* 2017, Buijs *et al.* 2018) we showed that such backpacks had only a very minor effect on hen behaviour after a two-day acclimation period (i.e., a slightly increased rate of pecking at their equipment only).

The 84 focal hens were selected at the start of the first monitoring cycle. Aiming to include hens that diverged in the time they spent on the range, 7 hens were caught inside and 7 hens were caught outside each house. In both locations, a group of hens was corralled into a frame enclosure and 7 hens without plumage damage or keel bone fractures were randomly selected. Fractures were assessed by palpation by a highly experienced assessor. Damaged individuals were excluded because the development of plumage and keel bone damage were indicators of interest in a different part of the study. Each focal hen was equipped with a backpack and three colour coded leg rings on each leg (to allow individual recognition from a distance). The neck feathers were trimmed slightly at the ends to minimise obscuring of the monitoring device. At the end of each monitoring cycle, the backpacks were removed to download the data.

Monitoring devices placed in the environment. Six monitoring devices were placed inside and two outside each house (Figure 1). The devices in the slatted area were cable-tied to the feeder or perch. The devices in the litter area and on the range were attached to plastic stakes which were pressed into the ground. Indoor devices were positioned where sunlight would come into the house at different times of the day (based on pilot observations). All devices were attached slightly above hen height (to avoid blocking of the sensors by passing hens) and at 1 m from the wall, except the devices attached to the feeder which were placed at the hens' chest level and 0.5 m from the wall. The missing data from each 17th minute of the ambient devices was replaced by the data of the 16th minute, as ambient light conditions were assumed not to differ greatly from one minute to the next.

Monitoring cycles and data processing. The system was set up and used in five (7-8 day long) cycles, starting when the hens were 20, 26, 32, 36 and 41 weeks of age (July-December 2017). On each day, monitoring started when the popholes were opened to allow access to the range (the devices nearest to the popholes were illuminated after pophole opening, allowing us to determine this time exactly).

Because hens were locked in after dark illumination of these devices could not be used to determine the end of range access. Instead, monitoring ended when the lowest level measured by either of the devices mounted outside exceeded the highest light level on the devices placed inside the house by less than 10% (excluding the devices placed near the popholes of the slatted area). A difference between the minimum outdoor reading and the maximum indoor reading of less than 10% also occurred occasionally during daytime, when sunlight was at the particular angle to shine through the ventilation slits onto the devices inside. Data from such periods was discarded, as a lack of difference between indoor and outdoor light levels was expected to cause errors when determining hens' location.

At the end of each monitoring cycle the data from all devices was downloaded. Each reading of a hen-mounted device that was recorded whilst the system was considered active (i.e., popholes open and an indoor-outdoor light difference ≥10%) was compared to the maximum indoor reading in the relevant house for that minute. Readings exceeding the maximum indoor value were used to classify the hen as outside, whereas readings below or equal to the maximum indoor value were used to classify the hen as inside during any particular minute.

Direct observation of hen location. Each of the 84 focal hens was observed directly for five minutes in each of the five monitoring cycles. Observation days started at 10 AM (to avoid the egg laying period) and ended between 4 and 8 PM depending on the season. During these five minutes the location of the hen (inside or outside) was recorded continuously using Obansys software (Mangold International, Arnstorf, Germany) on a tablet computer. When the hen was observed outside, it was also noted when she was in a clearly delineated shadow or in the pig shelter. Observations were conducted by three observers over the course of three days within each monitoring cycle. Focal hens were observed in a pre-determined order to avoid confounding between treatment/flock/individual and time of day.

Statistical analysis

Hen location as determined by monitoring (i.e., inside or outside, scored at 1-minute intervals) was compared to the hen's location as observed at the exact same time. Hen location as observed was considered the gold standard. From this comparison the accuracy (i.e., the percentage correctly classified), sensitivity (percentage classified as outside when truly outside) and specificity (percentage classified as inside when truly inside) were calculated per hen per monitoring cycle and then averaged per house per monitoring cycle. Accuracy and sensitivity were subsequently analysed in a linear mixed model with treatment (more vs. fewer structures), monitoring cycle (2-5) and their interaction as fixed factors and house as a random factor. Specificity was analysed using exact Wilcoxon rank sum tests to assess the effect of structures within each cycle because of clearly non-normal residuals.

The percentage of time spent outside as indicated by the monitoring system was analysed in a (binomial) generalized linear mixed model with treatment (more vs. fewer structures), cycle (2-5, categorical), original catching location (in vs. out) and their interactions as fixed factors, and house and hen as random factors. A sequential Bonferroni correction (Hochberg 1988) was applied to pairwise comparisons between cycles.

Correlation in individual range use over the observation cycles was evaluated using Spearman rank correlations.

All analyses were performed in R 3.3.3 (R Core Team, 2017), using the lme4, car, lsmeansLT, ggpubr, lmerTest, FSA and coin packages. Fixed effects with a P-value ≥ 0.10 were removed from the models.

Results

Data exclusion

All data from cycle one (July) was discarded because the ambient devices reached their maximum almost continuously, precluding determination of location based on a comparison of light levels. This problem did not persist in later cycles when light levels were lower (August-December).

In addition to data collected when the hens did not have access to the range (i.e., at night) some data from observation cycles 2-5 had to be discarded because light conditions inside and outside were too similar (1.6%), because no reading was acquired in the 17th minute (6%), because a hen-mounted device failed to record in that cycle (4×), or because a backpack strap snapped (1×). Three of the 84 focal hens died (one before and one during cycle 2, and one before cycle 4) and one hen could not be found when fitting equipment for cycle 2 but was equipped in later cycles. In all cases, the data is reported as a percentage of the non-discarded data.

Accuracy, sensitivity and specificity of the light-based monitoring system

Accuracy (i.e., the percentage of agreement between the monitoring system and direct observations) was high (Figure 4), at least 85% under all circumstances. In cycles two and four accuracy was (or tended to be) significantly higher when fewer structures were present on the range, whilst in cycle five the opposite was observed (structure × cycle interaction $F_{3,16}$=4.1, P=0.024). Rather than a true inversion of the effect of structures, this was likely due to closure of several popholes of one house during the last days of cycle five. This led to a considerable number of false positives as the indoor devices were no longer in the brightest places within the shed. After excluding this day for this house from the analysis accuracy was instead affected by main effects of structures ($F_{1,19}$=5.2, P=0.034, Least Squares Means (LSMEANS) more: 92%, fewer: 96%, SEM: 1) and cycle ($F_{3,19}$=3.2, P=0.047, LSMEANS: 89, 94, 95, 98% for cycles 2-5, respectively, SEM: 1). Accuracy in cycle 2 tended to be lower than in cycles 3 and 4 (P=0.092 and 0.066, respectively) and was significantly lower than in cycle 5 (P=0.007).

Sensitivity was also high, exceeding 80% under all circumstances and significantly higher in monitoring cycles 4 and 5 than in monitoring cycle 2 ($F_{3,20}$=3.5, P=0.036, Figure 4). No significant effect of structures ($F_{1,19}$=1.6, P=0.228) or a structure ×

cycle interaction ($F_{3,16}$=1.4, P=0.287) were found. These results were not affected by the exclusion of the data affected by pophole closure, as sensitivity was 100% in this house both before and after removal.

Specificity was often perfect (17 out of 24 house × monitoring cycle combinations), exceeding 90% under all circumstances. No significant effect of structures was found within any cycle (P≥0.4, Z=-1 - 1.2). Re-analysis after excluding the data affected by pophole closure led to similar conclusions. Overall specificity was 93% (± 18 SD), or 96 (10 ± SD) after data exclusion.

Sources of error

Notes made during the behavioural observations were used to identify possible sources of error. After data exclusion 1454 datapoints were left. Of these, 24 were false positives (hens classified as outside whilst truly inside) and 79 false negatives (hens classified as inside whilst truly outside). False positives most often occurred when the hen was near the pophole (15×), and in house six on the days that several popholes remained shut (11×). False negatives occurred when the hen was outside but in the shadow (24×), in hens whose neck feathers occasionally covered the sensor (11×), in the pig shelter (9×), and when dustbathing in a deep pit (3×). Both types of error occurred directly before and after the hen entered or exited the house (8×). For the other errors (3× false positive and 26× false negative) no likely reason could be identified.

Time spent outside as indicated by the light-monitoring devices

Hens from ranges with more structures tended to spend a greater percentage of time outside (χ^2_1=2.9, P=0.089, back transformed LSMEANS more structures: 67%, fewer structures: 56%, SEM: 4). Hens that had originally been caught outside spent a significantly greater percentage of time outside than those caught inside (χ^2_1=10.0, P=0.002, caught outside: 72%, caught inside: 51%, SEM: 4). Hens spent a significantly greater percentage of time outside in cycles 3 and 4 than in cycles 2 and 5 (χ^2_3=23.5, P<0.0001, cycle 2: 40%, cycle 3: 74%, cycle 4: 76%, cycle 5: 56%, SEM: 6). Pairwise differences between cycles were significant (P<0.05), except for cycle 3 vs. 5 (P=0.069). Removing the data from the last days of cycle 5 in the house where several popholes remained shut did not alter these results substantively.

The percentage of time individuals spent outside was significantly correlated between all monitoring cycles (P<0.0001). Stronger correlations occurred between cycles that were closer together in time (Figure 5). Again, removing the last days of cycle 5 in house 6 did not alter these results substantively.

Discussion

Comparison between monitoring data and direct observations by a human observer showed that our light-based system assessed range use very accurately (92-96%). Accuracy was only slightly below that of RFID systems that require narrow, tunnel-like popholes (97-98%, Thurner *et al.* 2010). Such systems are difficult to apply on a commercial farm without constraining range use by altering pophole space, number and location. The ease with which the static components of our light-based system can be set up (<30 minutes/house) as well as its flexibility (ambient device placement can be customized easily for each house) make it much more suitable for application in an on-farm setting. Accuracy increased throughout the experiment, probably due to modifications limiting daylight infiltration into the house (although other factors, e.g., changes in behaviour with age or season cannot be excluded fully). This suggests that if the system is used to compare houses that differ in their ingress of daylight, it will be necessary to check if accuracy is affected and if so, whether this results in a systematic over- or underestimation of ranging. Also, the relatively high number of false positives in one house when the popholes remained shut on one side emphasizes the importance of the correct placement of the ambient monitoring devices, at least one of which needs to be in the most brightly lit part of the house all the time.

More false negatives (classification as inside, whilst truly outside) than false positives (classification as outside, whilst truly inside) occurred, meaning that the system very slightly underestimated range use. Some of these false negatives seemed due to the hen being in a relatively dark outdoor area (e.g., in the shade, pig shelter or a dustbathing pit). However, hens were often in shaded areas without being misclassified, suggesting that it was a combination of shade and other factors that resulted in false negatives. Similarly, hens whose neck feathers were occasionally observed to cover the light monitoring device were responsible for a relatively high number of false negatives, but were often classified correctly when outside. Specific body postures may have resulted in feathers covering the device occasionally (even though feathers had been trimmed back). In addition, both false negatives and false positives occurred when hens exited or entered the house. This likely reflects delayed or pre-emptive scoring by the observer or a slight mismatch in the timers of the hen-mounted device and the tablet computer used for the observations. It should be emphasised that false positives and negatives represent a small proportion of the overall data collected.

We monitored specific focal hens in a predetermined order whilst they moved around the house and range. Theoretically, more data on e.g. the effect of shade could have been obtained by instead selecting hens from shaded and unshaded areas systematically. However, this would mean that the accuracy obtained would no longer reflect the accuracy as a whole, because this is determined by the time hens spend in different locations. For instance, hens' presence in the pig shelter always resulted in an incorrect classification, but this had almost no impact on overall accuracy as hens rarely used it. Although accuracy was slightly higher for ranges with fewer structures, we found no clear indication that this was specifically due to an increased number of false negatives as a result of more shaded outside areas, as the amount of structures was not found to affect sensitivity. This is likely also influenced by the type of structures we used, most of which were made of wind breaking cloth which only results in partial shading. In contrast, hens that were in the pig shelter (which provided full shade) were always misclassified as inside. Whether the pig shelter should be classified as an indoor or outdoor area is debatable however, and in any case hens spent little time in there.

Although we observed individual hens directly for a limited amount of time (5 min/hen/cycle) it needs to be emphasized that direct observation was only used to validate the light monitoring system, not to draw specific conclusions about individual hens. As such, we had over 3 hours of direct observation time per cycle per type of range (more vs. fewer structures), which we chose to spread over a high number of hens to minimize the chance that the results on accuracy were distorted by individuals with highly divergent behaviour. In contrast, analyses of the effects on range use were entirely based on the data obtained from the monitoring devices. Therefore, several days of data were available per hen per cycle, rather than 5 minutes. The percentage of hens per flock that was equipped was relatively low (0.7%), as we used a novel way of attaching the equipment to the hens which necessitated regular inspection of all focal hens for signs of discomfort, abrasion or damage to the equipment. This precaution prohibited us from equipping a larger proportion of the flocks. Although equipping more hens may be preferable in the future, the sample size used in the current study was sufficient to confirm our a-priori hypotheses.

The monitoring system was sensitive enough to detect a tendency for greater use of the ranges with more structures compared with fewer structures. In fact, the difference we found (67 vs. 56% of the monitored time spent outside) is more pronounced than indicated by previous research (Hegelund *et al.* 2005: 2% extra hens outside, Zeltner and Hirt 2008: 7% extra hens outside, Bestman and Wagenaar 2003: 2% extra hens outside for each 10% increment of range where cover was available for all-female 2000 hen flocks). Several other studies found no significant influence of structures on range use, although the structures did influence the distribution of the hens over the range (Gilani *et al.* 2014; Zeltner and Hirt, 2003; Pettersson *et al.* 2017). Differences between studies with respect to structure type, number, diversity and distance from the house probably contribute to the differences in the results. Additionally, all previous studies used direct observations, which may be prone to underestimating range use if the hens can hide from view behind or underneath the structures.

The system was also sensitive enough to detect that hens caught outside prior to the experiment ranged substantially more throughout the experiment than those that had been caught inside (72 vs. 51%). This supports the suggestion of the existence of clear individual differences in ranging behaviour within flocks, even when all hens within that flock are subjected to the same environment. Previous research has suggested two main categories of underlying reasons for such individual differences: biological predisposition (e.g., fear levels or exploratory tendencies: Campbell *et al.* 2016; Hartcher *et al.* 2016) and unequal ease of access (e.g., hens habitually roosting further from pop-holes thus having to perform more effort to go out, or hens less able to jump out of elevated popholes due to injury: Pettersson *et al.* 2018; Richards *et al.* 2012). The current study does not distinguish between these two possible explanations conclusively. However, it should be noted that the housing system used provided ease of access for all hens, as there were no elevated tiers, the stocking density was low, many large non-elevated popholes were available, and no roosting position was far from a pophole due to the rectangular house. The occurrence of substantial individual differences in range use, even when all hens should theoretically have had easy access to the range, indirectly supports the theory on biological predisposition as a driver for range use. Furthermore, it emphasizes the value of individual measurements rather than flock level estimates. The observed difference between hens originally caught inside and outside also suggests that hens' ranging habits are established at an early age, which may aid in the selection of hens with different profiles in future studies. Individual range use was also significantly correlated between all monitoring cycles. This association was strong for adjacent cycles (r_s^2=0.5-0.7), and somewhat weaker for those one or more cycles apart (r_s^2=0.4-0.5 and 0.2, respectively). This shows that although hens clearly form ranging habits, with some birds consistently spending more time outside than others, these habits may drift over time. Future research will be necessary to determine the reasons for such changes in range use.

To a certain extent, the difference in the percentage of time that hens originally caught inside and outside spent on the range also explains why range use was relatively high in our study. Our focal hens were collected equally inside and out, and because less than half of the flock was outside during selection this meant that hens with a high ranging tendency were overrepresented in our sample. However, even the focal hens that were caught indoors spent 51% of the monitored time outside. This is much higher than previously reported levels of range use in commercial flocks obtained by estimating number of hens on the range at any given time, which is a proxy for the percentage of time hens spent outside (Pettersson *et al.* 2017: 10%; Chielo *et al.* 2016: 13%; Gilani *et al.* 2014: 13%; Hegelund *et al.* 2005: 9%). However, previous studies using RFID technology also report a large percentage of time spent outside (Campbell *et al.* 2017b: 3-5 hours per day; Hartcher *et al.* 2016: 6 hours per day). This discrepancy between studies using an estimated number of hens outside and automated monitoring has been suggested to be due to the fact that RFID systems were used to study small experimental flocks (which usually range more, Pettersson *et al.* 2016). However, this suggestion is not in line with the high levels of range use we found in the present study, in which flocks of 2000 hens were used, which are representative for commercial organic egg production. Previous research has indicated that the percentage of hens outside is independent of flock size for flocks ≥ 2000 hens (Gebhardt-Henrich *et al.* 2014). Instead, differences between the present study and previous studies relying on counting the number of hens outside may be due to an underestimation of range use when counting birds. Such estimates collected alongside the current study suggested that on average less than 20% of the hens were outside during scans performed between 10 am and 3 pm, a much lower percentage than shown by automated monitoring. The underestimation in the flock level range use may be due to incomplete detection of all hens on large ranges, or the absence of observations during peak ranging times in the early morning and late evening (Bubier and Bradshaw, 1998; Dawkins *et al.* 2003).

In addition to a large percentage of time spent outside, we also found that all of our focal hens spent at least some time outside in each cycle. This contrasts with previous reports that some hens never venture out (Campbell *et al.* 2017b: 2%, Richards *et al.* 2011: 8%; Gebhardt-Henrich *et al.* 2014: 30%). The ease of access to the outdoor area may have contributed to this. Our hens did not have to navigate a high threshold to access the range, whereas in Richards *et al.* 2012 a 45 cm high barrier had to be crossed. Furthermore, in previous studies walking distances between the feeders and the outdoor area were often longer because a wintergarden or litter area had to be traversed (Gebhardt-Henrich *et al.* 2014; Richards *et al.* 2012), whereas in our study the nearest feeder was only 50 cm from an exit to the range. Also, indoor stocking density was lower and more and larger popholes were available in our study than in previous ones (Campbell *et al.* 2017a,b; Richards *et al.* 2012) making it less likely that a hen would be blocked on her way out. Favourable weather may also have contributed: during most cycles it was generally dry and mild, which stimulates range use (Chielo *et al.* 2016; Gilani *et al.* 2014). In contrast, during the last cycle it was relatively cold, wet and windy, and range use was 20% lower than in the preceding cycle. Until that time, range use had increased progressively with age, in line with previous reports (Campbell *et al.* 2017b). However, as increases in age coincided with changes in weather patterns it is not entirely clear which of these two factors altered range use in the current and previous studies (Richards *et al.*, 2012; Hegelund *et al.*, 2005).

In conclusion, our light-based system monitored range use accurately, with high sensitivity and specificity. Accuracy was only slightly influenced by levels of range cover. The system's performance compared favourably with RFID systems that need to cover the full length of each access point and therefore either require specific modifications to range access (which in themselves may influence ranging behaviour), or a large amount of equipment. The light monitoring system works independently of the number and size of access point and only requires small devices that can be set up quickly in a flexible manner to measure range use in a variety of housing systems. It is therefore highly suitable for use on commercial farms. However, houses which allow more daylight to enter, and fully shaded areas on the range, may decrease the system's accuracy. The extent of this decrease will depend on how often these areas are used by the hens. Hens were shown to have relatively consistent ranging habits that can already be predicted two weeks after they are first allowed to access the range. Further research is required to determine the cause of these consistent individual differences in range use.

Acknowledgement

The authors acknowledge BBSRC for funding this project, as well as support from Stonegate.

Declaration of interest

There are no conflicts of interest to declare.

Ethics statement

The study was carried out following ethical approval by the University of Bristol.

Software and data repository resources

None of the data or models were deposited in an official repository.

References

Bestman MWP and Wagenaar JP 2003. Farm level factors associated with feather pecking in organic laying hens. Livestock Production Science 80, 133-140.

Bubier NE and Bradshaw RH 1998. Movement of flocks of laying hens in and out of the hen house in four free range systems. British Poultry Science 39, S5-S18.

Buijs S, Nicol CJ, Booth F, Richards G and Tarlton J 2017. Effects of automated monitoring equipment on laying hen behaviour. In Proceedings of the 7th International Conference on the Assessment of Animal Welfare at Farm and Group Level. 5–8 September 2017, Ede, The Netherlands, p. 116.

Buijs S, Booth F, Richards G, McGaughey L, Nicol CJ, Edgar J and Tarlton JF 2018. Behavioural and physiological responses of laying hens to automated monitoring equipment. Applied Animal Behaviour Science 199, 17-23.

Campbell DLM, Hinch FN, Downing JA and Lee 2016. Fear and coping styles of outdoor-preferring, moderate-outdoor and indoor-preferring free-range laying hens. Applied Animal Behaviour Science 185, 73-77.

Campbell DLM, Hinch FN, Downing JA and Lee 2017a. Outdoor stocking density in free-range laying hens: effects on behaviour and welfare. Animal 11, 1036-1045.

Campbell DLM, Hinch FN, Dyall TR, Warin L, Little BA, Downing JA and Lee 2017b. Outdoor stocking density in free-range laying hens: radio-frequency identification of impacts on range use. Animal 11, 121-130.

Chielo LI, Pike T and Cooper J 2016. Ranging behaviour of commercial free-range laying hens. Animals 6, article number 28.

Dawkins MS, Cook PA, Whittingham MJ, Mansell KA and Harper AE 2003. What makes free-range broiler chickens range? In situ measurement of habitat preference. Animal Behaviour 66, 151-160.

Gebhardt-Henrich SG, Toscano MJ and Frohlich EKF 2014. Use of outdoor ranges by laying hens in different sized flocks. Applied Animal Behaviour Science 155, 74-81.

Gilani AM, Knowles TG and Nicol CJ 2014. Factors affecting ranging behaviour in young and adult laying hens. British Poultry Science 55, 127-135.

Hartcher KM, Hickey KA, Hemsworth PH, Cronin GM, Wilkinson SJ and Singh M 2016. Relationship between range access as monitored by radio frequency identification technology, fearfulness, and plumage damage in free-ranging laying hens. Animal 10, 847-853.

Hegelund L, Sorensen JT, Kjaer JB and Kristensen IS 2005. Use of the range area in organic egg production systems: effect of climatic factors, flock size age and artificial cover. British Poultry Science 46, 1-8.

Hochberg Y 1988. A sharper Bonferroni procedure for multiple tests of significance. Biometrika 75: 800-803.

Knierim U 2006. Animal welfare aspects of outdoor runs for laying hens: a review. NJAS-Wageningen Journal of Life Sciences 54, 133-145.

Lambton SL, Knowles TG, Yorke C and Nicol CJ 2010. The risk factors affecting the development of gentle and severe feather pecking in loose housed laying hens. Applied Animal Behaviour Science 123, 32-42.

Larsen H, Cronin G, Smith CL, Hemsworth P and Rault J-L 2017. Behaviour of free-range laying hens in distinct outdoor environments. Animal Welfare 26, 255-264.

Lindholm C, Karlsson L, Johansson A and Altimiras J 2016. Higher fear of predators does not decrease outdoor range use in free-range Rowan Ranger broiler chickens. Acta Agriculturae Scandinavica Section A – Animal Science 66, 231-238.

Mahboub HDH, Muller J and Von Borell E 2004. Outdoor use, tonic immobility, heterophil/lymphocyte ratio and feather condition in free-range laying hens of different genotype. British Poultry Science 45, 738-744.

Nagle TAD and Glatz PC 2012. Free range hens use the range more when the outdoor environment is enriched. Asian-Australian Journal of Animal Science 25, 584-591.

Nicol CJ, Potzsch C, Lewis K and Green LE 2003. Matched concurrent case-control study of risk factors for feather pecking in hens on free-range commercial farms in the UK. British Poultry Science 44, 515-523.

Pettersson IC, Freire R and Nicol CJ 2016. Factors affecting ranging behaviour in commercial free-range hens. World’s Poultry Science Journal 72, 137-150.

Pettersson IC, Weeks CA and Nicol CJ 2017. Provision of a resource package reduces feather pecking and improves ranging distribution on free-range layer farms. Applied Animal Behaviour Science 195, 60-66.

Pettersson IC, Weeks CA Norman KI and Nicol CJ 2018. Internal roosting location is associated with differential use of the outdoor range by free-range laying hens. British Poultry Science 59, 135-140.

R Core Team 2017. R: A language and environment for statistical computing, https://www.R-project.org/. R Foundation for Statistical Computing, Vienna, Austria.

Richards GJ, Wilkins LJ, Knowles TG, Booth F, Toscano MJ, Nicol CJ and Brown SN 2011. Continuous monitoring of pophole usage by commercial housed free-range hens throughout the production cycle. Veterinary Record 169, 338-341.

Richards GJ, Wilkins LJ, Knowles TG, Booth F, Toscano MJ, Nicol CJ and Brown SN 2012. Pophole use by hens with different keel fracture status monitored throughout the laying period. Veterinary Record 170, 494-498.

Stadig LM, Ampe B, Rodenburg TB, Reubens B, Maselyne J, Zhuang S, Criel J and Tuyttens FAM 2018. An automated positioning system for monitoring chickens’ location: Accuracy and registration success in a free-rang area. Applied Animal Behaviour Science 201, 31-39.

Thurner S and Wendl G 2005. Individual ranging behaviour of laying hens. Landtechnik 60, 30-31.

Thurner S, Maier S, Icken W, Wendl G and Preisinger R 2010. Identification reliability of laying hens at the wide electronic pophole. Landtechnik 65, 139-141.

Zeltner E and Hirt H 2003. Effect of artificial structuring on the use of laying hen runs in a free-range system. British Poultry Science 44, 533-537.

Zeltner E and Hirt H 2008. Factors involved in the improvement of the use of hen runs. Applied Animal Behaviour Science 114, 395-408.

List of figure captions

Figure 1. Schematic overview of the laying hen house (9 × 45 m) and placement of the ambient light measuring devices. The houses were windowless, but natural light could enter the house trough adjustable ventilation openings running along the full length of both sides of each house and through the popholes (when opened).

Figure 2. Structures present on all ranges with laying hens: ① trampolines (4× per range), ② fir tree stack (1× per range), and on the 'more structures' ranges only: ③ pig shelters (2× per range), ④ tents (4× per range), ⑤ artificial zig-zag hedges (5× per range).

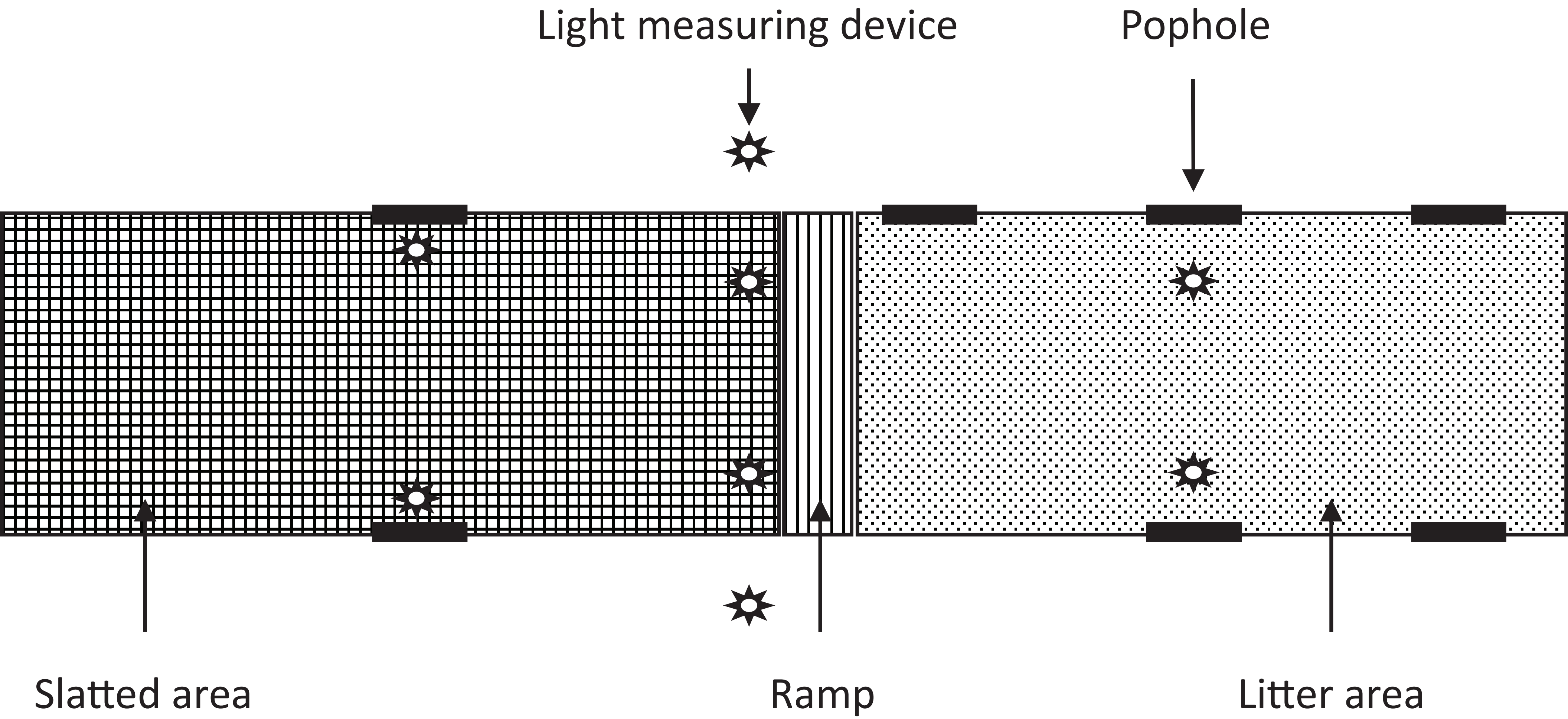
Figure 3. Laying hens fitted with equipment backpacks containing the light monitoring devices used to assess range use. In the picture on the left the arrow indicates the backpack, in the picture on the right it indicates the top of the light monitoring device sticking out of the wrapping. Photographs were taken in a different study, but the equipment and its attachment were identical.

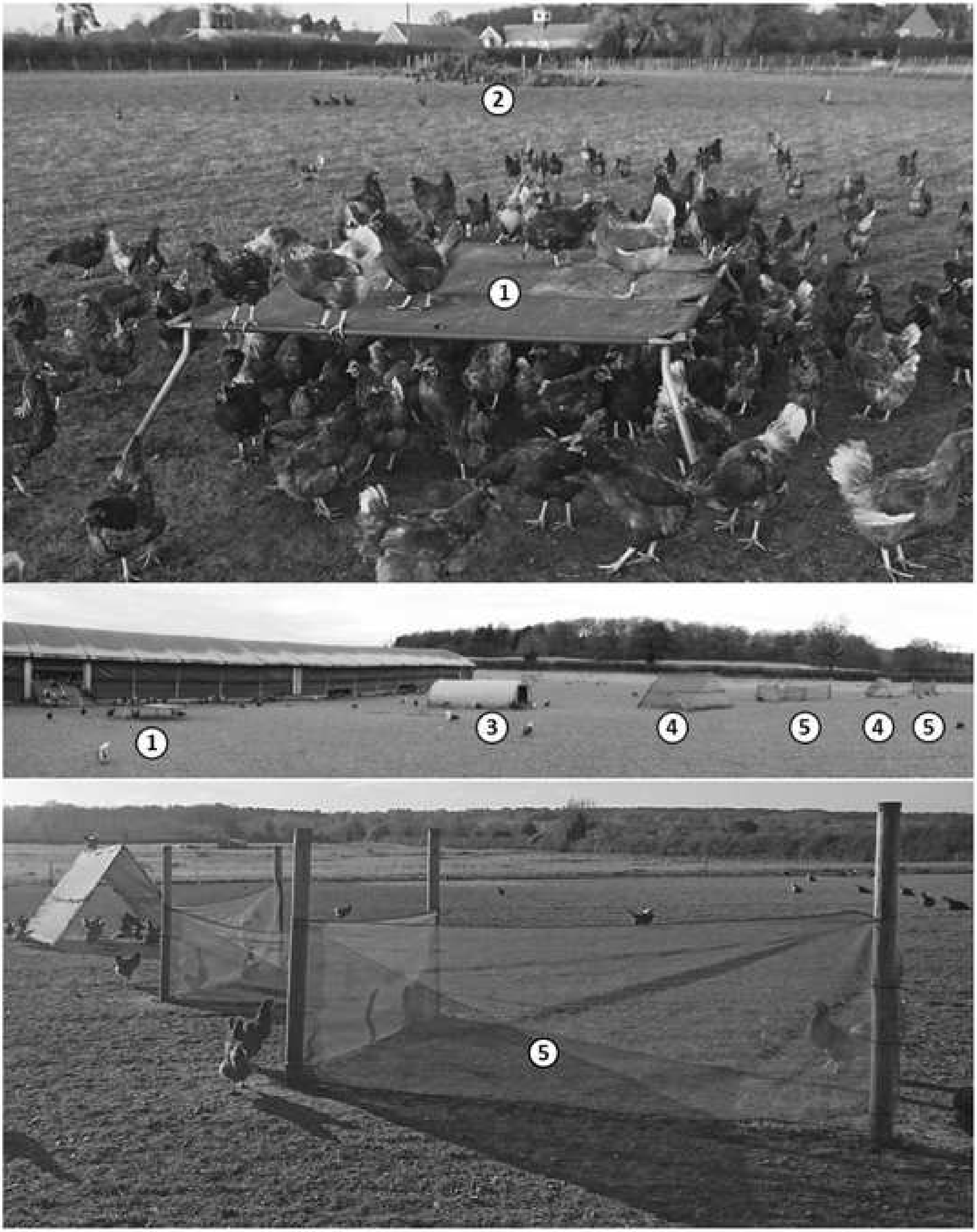
Figure 4. Accuracy and sensitivity of the system monitoring range use in laying hens. Note that data of one house where popholes were closed on one side during the last observation day is included (see text for values excluding these data). Exact values shown on bottom of bar.

*Significant difference between ranges with more and fewer structures within a cycle ($P<0.05$). [#]Tendency for a difference between ranges with more and fewer structures within a cycle (P<0.10). ns: no significant difference between ranges with more and fewer structures within a cycle (P>0.10). LSMEANS: Least Squares Means. LSMEANS lacking a common letter differ significantly (P<0.05) within treatment (accuracy) or overall (sensitivity).

Figure 5. Spearman correlations between the percentage of the monitored time that individual hens spent outside during the different cycles as indicated by the light monitoring devices. Squares: hens on ranges with more structures, circles: hens on ranges with fewer structures, grey: hens originally caught outside, black: hens originally caught inside. P<0.0001 for all correlations.

Figure 1 converted by EO




2
1
1
3
4
5
4
5
5

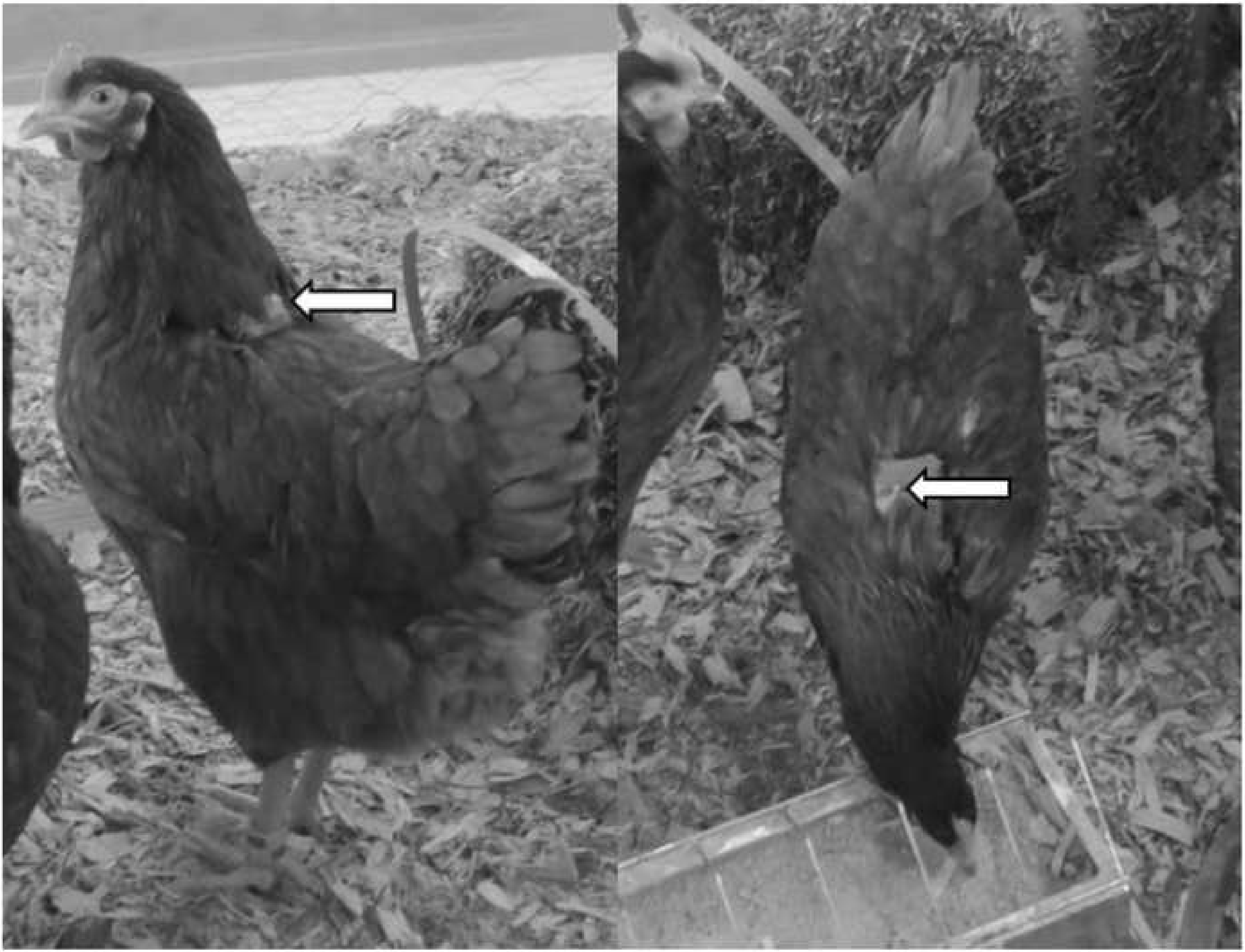


Figure 4 converted by EO

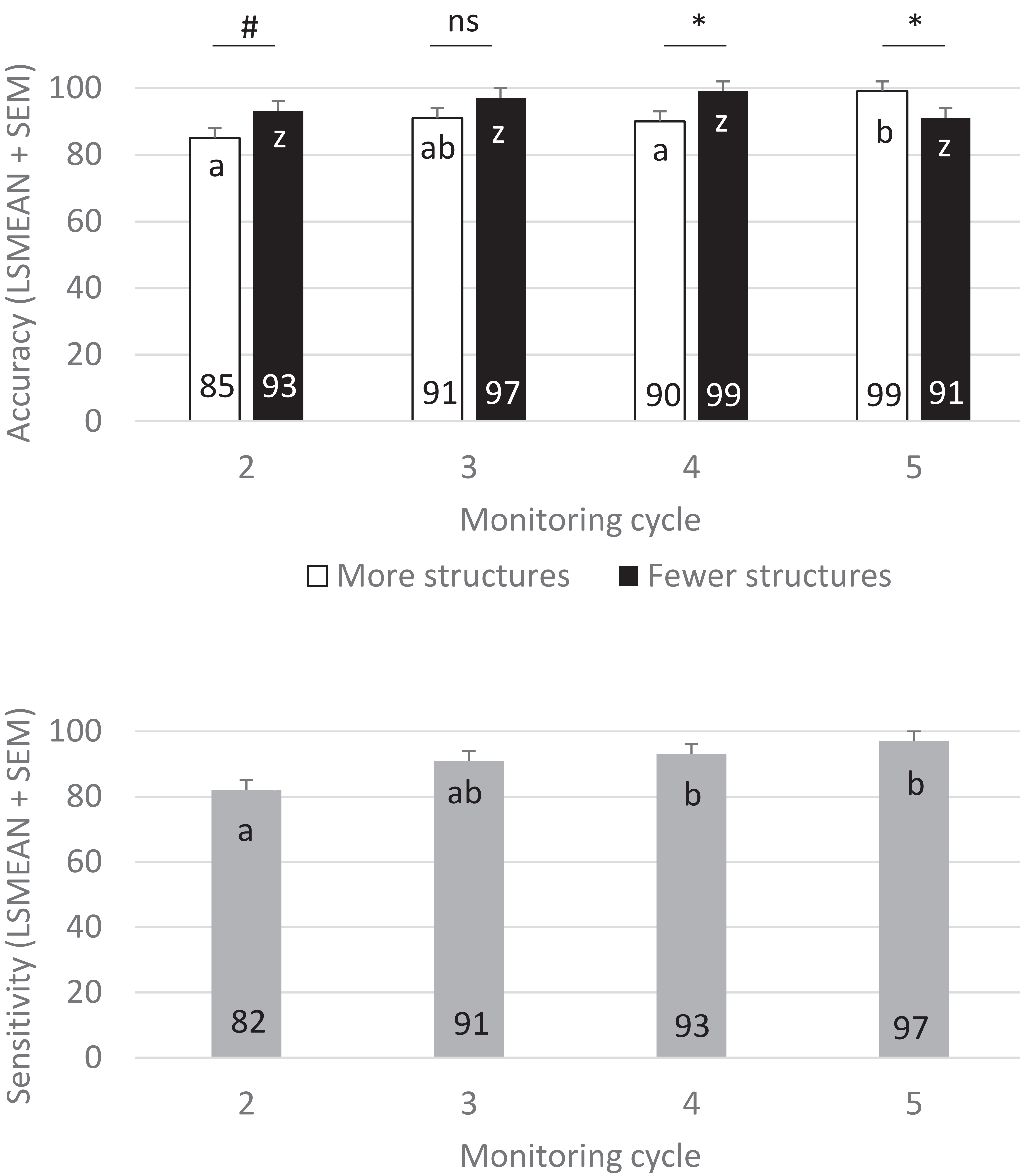
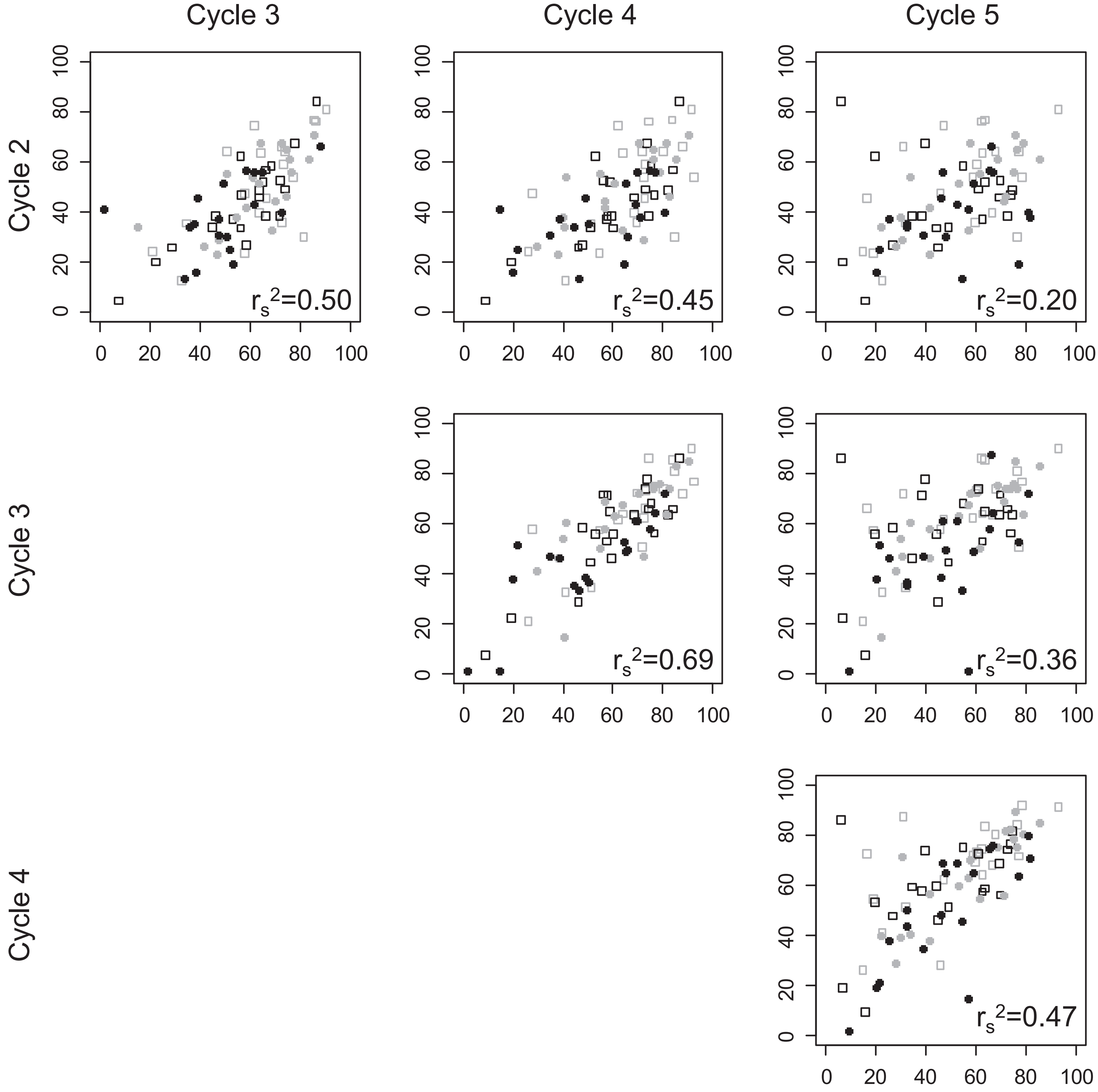


Figure 5



Cycle 3
Cycle 4
Cycle 5
Cycle 2
Cycle 3
Cycle 4
r_s^2=0.50
r_s^2=0.45
r_s^2=0.20
r_s^2=0.69
r_s^2=0.36
r_s^2=0.47

We have used all sections of the technical revisions checklist to review the document and made the following changes:

- An ethics statement, declaration of interest and repository statement were added.
- All references were checked for correspondence between in-text citations and the reference list. In some references Pettersson was misspelled. This is now rectified (all references now spelled with ss instead of s). Reference font size was changed in line 524. Hegelund 2005 was changed to Hegelund *et al.* 2005 (line 555).
- RFID and LSMEANS were defined at their first use.
- LSMEANS and ns were defined in the captions and the species was indicated in each caption

The figures were checked. However, a high-resolution file wasn't created for fig 2 and 4. We assume that this is because these are line drawings and therefore do not require a high resolution file. However, the text in figure 5 has become blotchy and more difficult to read during the conversion process. We are unsure how to remedy this though, as all text was sharp in the originally submitted version. Could you tell us how this could be solved?